INVESTIGATION AND MECHANISTIC STUDY OF SOME OF THE BIOACTIVE MOLECULES BY ELECTROCHEMICAL SENSORS: A VOLTAMMETRIC STUDY

Final Synopsis of the thesis to be submitted to

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DOCTOR OF PHILOSOPHY

IN

CHEMISTRY

By

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Introduction

Modern world is the world of sensors. Sensor is a device that detects the signals from its surroundings and converts it to meaningful information. Electrochemical sensors belong to class of chemical sensors where an electrode is used as a transducer element in the presence of an analyte. Modern electrochemical sensors are used to detect the various parameters in everyday life. In electrochemical sensor a receptor binds the analyte, a transducer to convert the reaction a measurable electrical signal. The electrode surface is used as the site for the reaction. The electrode will either reduce or oxidize the analyte of interest. The current produced during the reaction is monitored and used to calculate the concentration of the sample. In most of the electrochemical sensors, the electrode surface is modified with membranes, catalysts, or any other modifier to make the electrode more sensitive and more selective towards the electrolyte. Biosensors necessitate similar elements as electrochemical sensors in order to function, with a minor modification. The elements obligatory for biosensor is a receptor that binds the sample, an interface for the sample to react and a transducer. Biosensors play a major role in detective work of the levels of diverse substances in the human body. Sensors must be stable with little variation for a particular experiment, in addition to accuracy and precision in the data. Biosensors are specific in nature and the parameters measured must be confirmed by the experimenter.

It is observed that, the interest has been increased in the construction of modified electrode surfaces that differ from the corresponding bare surfaces and derive an electrode surface generating reproducible result, having the massive applications in biological and chemical sensing. The work in this thesis is focused to alter the properties of carbon surfaces controllably, so that the surface of the carbon can be utilized for desired sensor applications. As carbon is highly conducting with a wide potential window, comparatively inexpensive, structurally steady and stable layers of modifiers can attach the surface in a governable manner, carbon surface is chosen. Owing to the numerous application potentials of the modified electrodes, modification of the

electrode surface is of ultimate importance. The decorum of modification attained by conferring precise molecules accomplishes chemical specificity to the material.

A large number of researches have been focused on the applications of carbon paste electrodes for the fabrication of electrochemical sensors and biosensors [1-4]. Electrochemical approaches have become practical for the fabrication of electrodes [5-7]. Chemically modified electrodes based on different modifying material have attracted more attention because of their characteristics of low cost, easy operation, fast response, high sensitivity. The different materials such as amino acids, conducting polymer, metal nanoparticles, ionic liquids, self-assembled monolayer and multi-walled or single walled carbon nanotube have been applied in the modification of electrodes for the determination of different electroactive species [8-12].

In this work the Bare Carbon Paste electrode (BCPE) is derived by grinding the carbon material with a binder (w/w%) and Modified Carbon Paste electrode (MCPE) was prepared by electropolymerization, for dopamine, uric acid, ascorbic acid, tyrosine, vanillin, riboflavin and curcumin investigated electrochemically through voltammetric responses even in their interfering character [13-19]. Enormous modified carbon paste electrodes developed and have shown substantial enhancements of the electrochemical sensor towards the better performance of some bioactive molecules. The designed electrode represents easy preparation, quick reply, good stability and better sensitivity [20-21]. Electrodes treated with electropolymerized conducting polymer film have shown greater consideration in the past few decades, Studies have specified that polymer film modified electrodes display amplified current-voltage output for the detection of various bioactive molecules.

Thesis summary

The main focus of the effort in the work is to advance the carbon surface properties in order to develop the idea that the surface would be approachable to progress the desire sensor with

applications. In this proposed work carbon nanotube and graphite powder has been selected towards the study of the electrochemical properties of various bioactive molecules and the process that takes place across the electrolyte and the surface of the electrode. Carbon exhibits excellent conducting capacity with an extensive potential window, structural stability, comparatively inexpensive and the formation of an adherent stable layer of the modifier on the carbon surface. The current work designates the progress of the novel analytical technique for the recognition of different bioactive molecules. It also signifies the contribution of a new analytical method applicable to food, medicinal samples. The foremost intention of this work is to study the resourcefulness of the different CPEs with a unique modifier in terms of its applications to investigate the electrochemical behavior of different bioactive molecules and the sequence of emission of the electron transfer occurs across the electrode surface. The researcher believes that the contribution towards this work from the research laboratory is outlined in the work, may offer an exciting insight in the field of research work.

Organization of Thesis

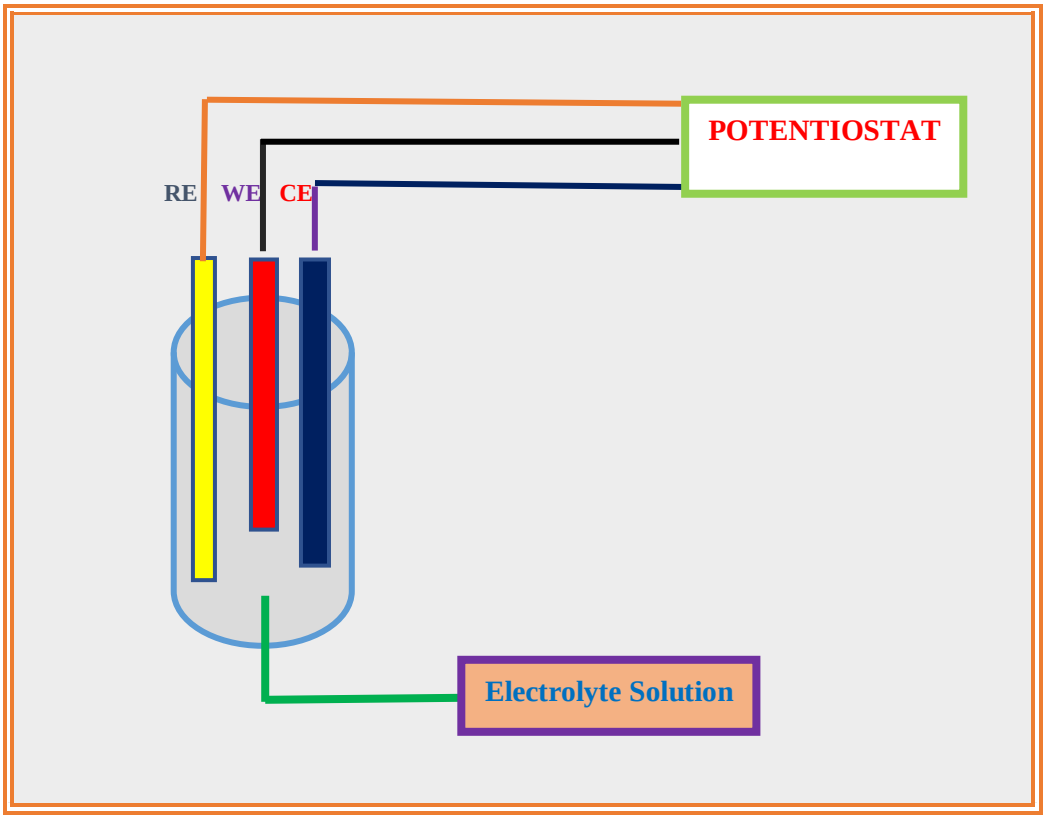
The entitled thesis "Investigation and Mechanistic Study of Some of the Bioactive Molecules by Electrochemical Sensors: A Voltammetric Study" has been divided into the following 8 chapters.

Chapter-1: Preface, Appraisal of Cyclic Voltammetry and Theoretical Discussions

This chapter includes the basic concepts of cyclic voltammerty such as introduction, techniques, theoretical aspects and applications of voltammetry. According to the literature survey, cyclic voltammetric investigation of some of the biologically active molecules is also been reviewed. This chapter also includes the scope and objective of the present work.

Chapter-2: Experimental

This chapter describes the concept, instruments used, the electrode system utilized, techniques followed and the procedures applied in the current work. Basically, the preparations of modified carbon developed electrodes are also discussed in this chapter. It contains the basic instruments required for the work such as potentiostat, a recording device like personal computer, and an electrochemical cell. A standard electrochemical cell consists of three electrodes immersed in an electrolytic solution of analyte. Three electrodes are nothing but the one is working electrode (WE) as bare and modified carbon paste electrodes, the reference electrode (RE) such as calomel electrode. The counter electrode (CE) is normally derived from an inert material.

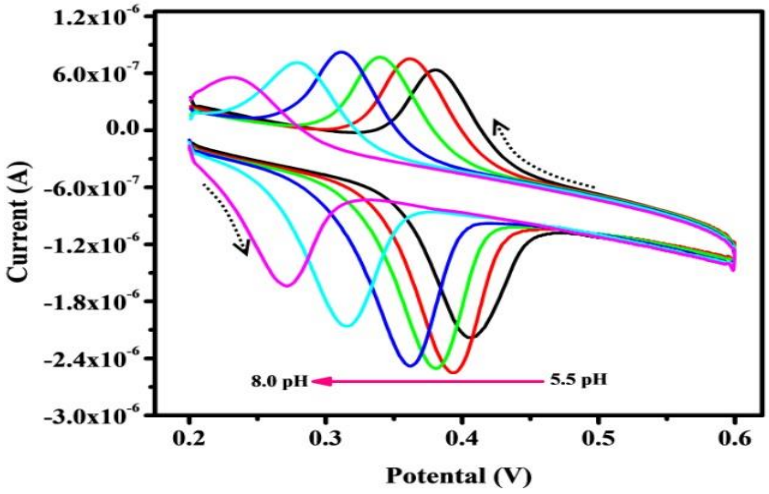


A Typical Electrochemical Cell

Chapter – 3: Electrochemical analysis of indigo carmine using polyarginine modified carbon paste electrode

(Journal of Electrochemical Science and Engineering, 11 (2), 2021, 87- 96, DOI:10.5599/jese.953, Citescore:1.7)

This chapter describes a suitable electrocatalytic method for the selective determination of indigo carmine (IC) at polyarginine modified carbon paste electrode (PAMCPE). The influence of different parameters such as IC concentration, solution pH and potential scan rate on the electrode responses is studied using cyclic and differential pulse voltammetry techniques. No interference is noticed at simultaneous presence of IC and riboflavin (RF) in the solution. The electrocatalytic current of IC at PAMPCE is varied linearly with its concentration in two separate ranges, from 0.2 μM to 1 μM, and 1.5 μM to 3.5 μM. Limits of detection and quantification are determined as 0.02 μM and 0.08 μM, respectively. The developed PAMCPE is showing successful reproducibility and stability. It is also found sensitive and reliable for trace amounts of IC in some real water and food samples. Since preparation of PAMCPE sensor is simple and easy, it could become a part of the standard method for determination of IC in real samples.

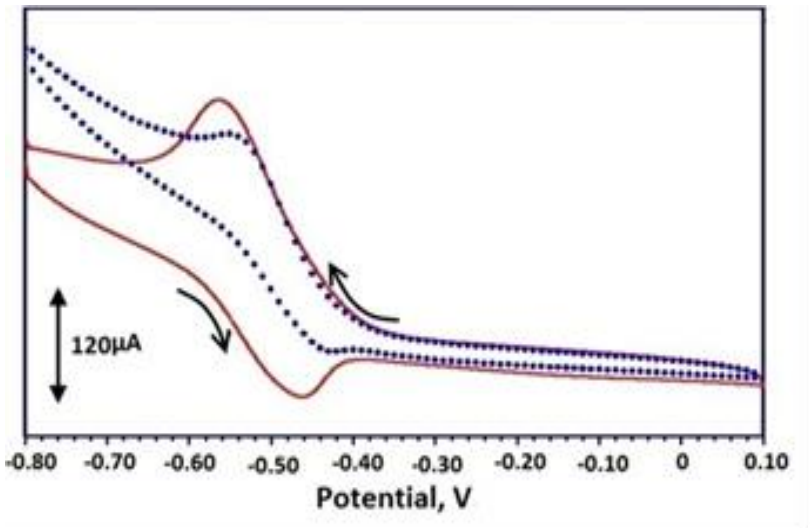


CVs of IC (10 μM) at PAMCPE in 0.1 M PBS at pH values from 5.5 to 8.0

Chapter 4: Polymer Modified Carbon Paste Electrode as a Sensitive Sensor for the Electrochemical Determination of Riboflavin and Its Application in Pharmaceutical and Biological Samples

(Analytical and Bioanalytical Chemistry Research, 7 (4), 2020, 461- 472, Citescore:2.6)

This chapter deals with the preparation of Poly (threonine) modified carbon paste electrode (PTMCPE) as a responsive and selective electrocatalytic sensor for the estimation of riboflavin (RF) by means of cyclic voltammetry (CV) and differential pulse voltammetry (DPV). The proposed electrode creates a catalytic enhancement in redox behavior of RF when equated with bare carbon paste electrode (BCPE). Under optimized conditions, RF quantification at modified electrode has shown a detection limit of 0.178 μM in the linear range of 0.2-1 μM and 1-5 μM. Simultaneous analysis of RF with folic acid (FA) and ascorbic acid (AA) shows well-separated peaks leading to a quick and selective analysis of RF. The fabricated sensor can be successfully applied to RF analysis in human blood serum and B- complex tablet with acceptable results.



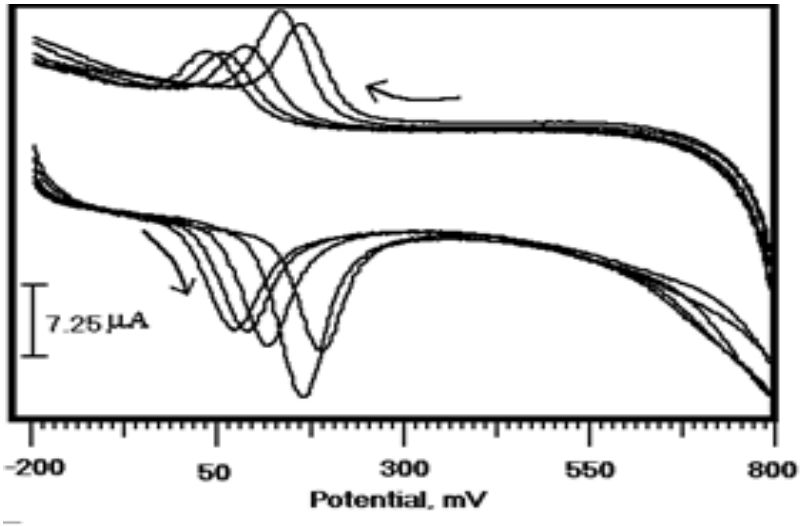
CV of RF at bare (dotted line) and PTMCPE (solid line) in 0.1 M PBS, 7.0 pH at the sweep rate of 0.1 V/s.

Chapter 5: Electrochemical Determination of Dopamine and Uric acid using Poly (proline) Modified Carbon Paste Electrode: A Cyclic Voltammetric Study

(Chemistry and Chemical Technology, 15 (2), 2021, 153-160, DOI:10.23939/chcht15.02.153, Citescore:1.2)

In this chapter a new method of cyclic voltammetric technique was used for electropolymerization of proline on the surface of carbon paste electrode and for individual and concurrent determination

of dopamine (DA) and uric acid (UA). The surface morphology of the developed electrode was studied by using field emission scanning electron microscopy. The modified electrode showed a high current response towards DA as compared to the bare electrode. The modified electrode shows good catalytic activity with a different oxidation potential of DA and UA. The electrode process was found to be adsorption controlled. The developed method shows very good stability and reproducibility. Under the optimized conditions, the concentration range is 0.01 μM to 0.02 μM and the observed detection limit was 4.7 μM. The developed sensor was applied for the determination of DA in the real sample with a good recovery.



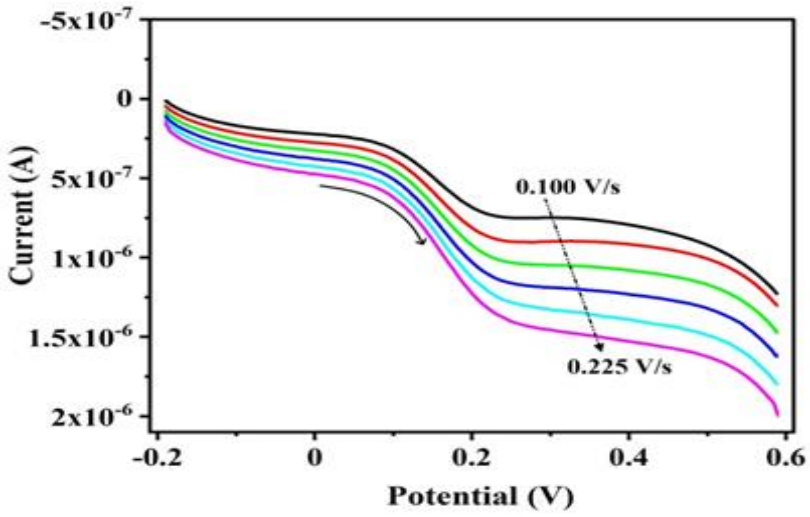
CVs of DA (0.01 μM) at PPMCPE in 0.2 M PBS at pH values of 6.0, 6.5, 7.0, 7.5 and 8.0

Chapter 6: Electro-polymerized Titan yellow Modified Carbon Paste Electrode for the Analysis of Curcumin

(Surfaces 4, 2021, 191–204. DOI:10.3390/surfaces4030017)

This chapter includes a modest, efficient, and sensitive chemically modified electrode was fabricated for sensing curcumin (CRC) through an electrochemically polymerized titan yellow (TY) modified carbon paste electrode (PTYMCPE) in phosphate buffer solution (pH 7.0). Cyclic voltammetry (CV) linear sweep voltammetry (LSV) and differential pulse voltammetry (DPV) approaches were used for CRC detection. PTYMCPE interaction with CRC suggests that the

electrode exhibits admirable electrochemical response as compared to bare carbon paste electrode (BCPE). Under the optimized circumstances, a linear response of the electrode was observed for CRC in the concentration range 2 μM to 10 μM with a limit of detection (LOD) of 1.094 μM. Moreover, the effort explains that the PTYMCPE electrode has a hopeful approach for the electrochemical resolution of biologically significant compounds. Additionally, the proposed electrode has demonstrated many advantages such as easy preparation, elevated sensitivity, stability, and enhanced catalytic activity, and can be successfully applied in real sample analysis.



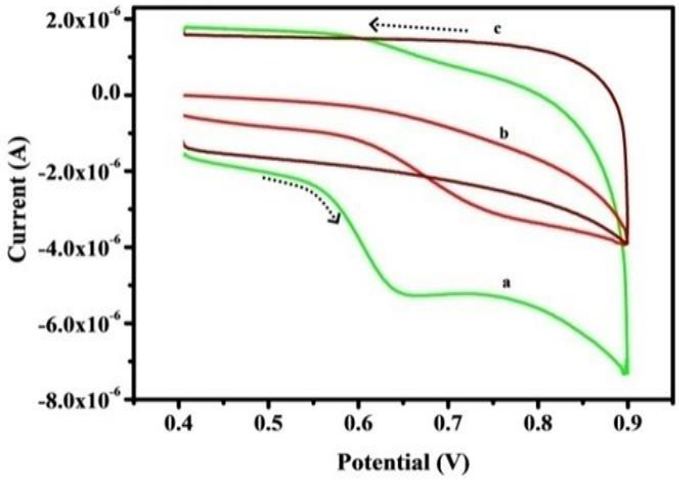
Linear Sweep Voltammogram of CRC (10 μM) at PTYMCPE in 0.1 M PBS of pH 6.5 at different scan rate (0.100 to 0.225 V/s)

Chapter 7: Rapid Electrochemical Monitoring of Tyrosine by Poly (Riboflavin) Modified Carbon Nanotube Paste Electrode as a Sensitive Sensor and Its Applications in Pharmaceutical Samples

(Biointerface Research in Applied Chemistry, 11(6), 2021, 14661-14672, Citescore:1.6)

In this chapter a poly(riboflavin) modified carbon nanotube paste electrode (PRFMCNTPE) is employed as a compatible and electrocatalytic sensor for the determination of Tyrosine (TYR). The analysis and assessment are carried out through Cyclic Voltammetry (CV) and Differential Pulse Voltammetry (DPV). The surface morphology of the intended sensor is studied all the way

through Field Emission Scanning Electron Microscopy (FE-SEM). The modified electrode shows outstanding electrocatalytic activity for TYR with high sensitivity and selectivity when compared to bare carbon nanotube paste electrode (BCNTPE). The electro-oxidation peak current of TYR and its concentration is found linear from 2 µM to 10 µM with a detection limit (LOD) of 0.45 µM. The developed sensor is productively applied for the determination of TYR in pharmaceutical samples like Tyrosine capsules. The adapted electrode shows good stability, exceptional reproducibility, and remarkable sensitivity.



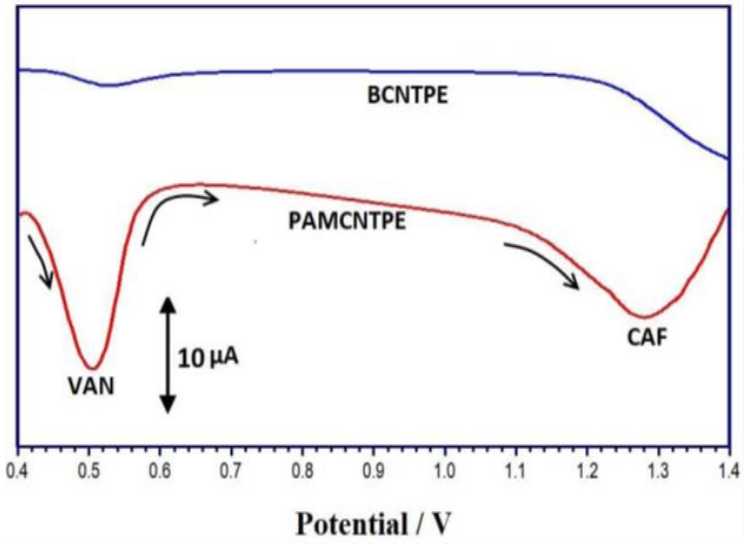
Cyclic Voltammogram of TYR at PRFMCNTPE (curve a), BCNTPE (curve b) and blank (curve c) in 0.1 M PBS, 7.0 pH at the sweep rate of 0.1V/s.

Chapter 8: Arginine Electropolymerized Carbon Nano tube paste electrode as Sensitive and Selective Sensor for Electrochemical Determination of Vanillin

(Journal of Materials and Environmental Sciences, 11 (3) 2020, 512-521)

This chapter describes the construction of Poly (Arginine) modified carbon nanotube paste electrode (PAMCNTPE) was prepared and it was approachable and discriminatory electrocatalytic sensor for Vanillin (VAN) determination using Linear Sweep Voltammetry (LSV), Cyclic Voltammetry (CV) and Differential Pulse Voltammetry (DPV). Under the optimized circumstances, the quantification of VAN at the developed electrode has revealed the linear range

2 μM to 10 μM and 15 μM to 45 μM and a limit of detection 0.033 μM. Instantaneous resolving of VAN and Caffeine (CAF) shows a well separated peaks at 0.508 V and 1.281 V. Applicability of the developed electrode is studied using food sample, shows the recovery in the range of 95.92% to 99.73%, this indicates that the modified electrode is having better analytical performance and can be used for the determination of VAN in commercial food samples.



DPVs of the solution containing VAN (100 μM) and CAF (100 μM) in 0.1 M PBS (pH 7.5) at the PAMCNTPE and BCNTPE

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List of publications: -

Papers Published/Accepted/Communicated in national/international journals

1. Edwin S. D'Souza, J. G. Manjunatha, C. Raril, G. Tigari, D. K. Ravishankar and H. J. Arpitha, Electrochemical analysis of indigo carmine using polyarginine modified carbon paste electrode, Journal of Electrochemical Science and Engineering, 11(2), 2021, 87-96, DOI:10.5599/jese.953. Citescore:1.7.

2. **Edwin S. D' Souza**, J. G. Manjunatha, C. Raril, G. Tigari and P. A. Pushpanjali, Polymer Modified Carbon Paste Electrode as a Sensitive Sensor for the Electrochemical Determination of

Riboflavin and Its Application in Pharmaceutical and Biological Samples, Analytical and Bioanalytical Chemistry Research, 7 (4), 2020, 461-472, Citescore:2.6.

3. Edwin S. D' Souza, J. G. Manjunatha, C. Raril, Electrochemical determination of dopamine and uric acid using poly (proline) modified carbon paste electrode: A cyclic voltammetric study. Chemistry and Chemical Technology, 15(2), 2021, 153–160 DOI:10.23939/chcht15.02.153, Citescore:1.2.

4. Edwin S. D'Souza, J. G. Manjunatha, C. Raril, G. Tigari, H. J. Arpitha, and S. Shenoy, Electro-Polymerized Titan Yellow Modified Carbon Paste Electrode for the Analysis of Curcumin, Surfaces 4, 2021, 191–204. DOI:10.3390/surfaces4030017.

5. **Edwin S. D' Souza**, J. G. Manjunatha, C. Raril, G. Tigari, and D. K. Ravishankar, Rapid Electrochemical Monitoring of Tyrosine by Poly (Riboflavin) Modified Carbon Nanotube Paste Electrode as a Sensitive Sensor and its Applications in Pharmaceutical Samples., Biointerface Research in Applied Chemistry, 11(6), 2021, 14661-14672, Citescore:1.6.

6. Edwin S. D' Souza, J. G. Manjunatha, C. Raril, G. Tigari, D. K. Ravishankar and Santosh Fattepur, Arginine Electropolymerized Carbon Nanotube Paste Electrode as Sensitive and Selective Sensor for Electrochemical Determination of Vanillin, Journal of Materials and Environmental Sciences, 11 (3), 2020, 512-521.

7. B. T. Harshitha, J. G. Manjunatha, P. A. Pushpanjali, C. S. Karthik, S. Sandeep, P. Mallu, **Edwin S. D'Souza,** N, Sreeharsha, S. M. B. Asdaq, and M. K. Anwer, Efficient Electrochemical Determination of Catechol with Hydroquinone at Poly (L-Serine) Layered Carbon Paste Electrode. ChemistrySelect 6, 2021, 6764 –6772, DOI:10.1002/slct.202101809, IF:2.109.

8. P. A. Pushpanjali, J. G. Manjunatha, N. Hareesha, **Edwin S. D' Souza,** M. M. Charithra, and N. S. Prinith, Voltammetric analysis of antihistamine drug cetirizine and paracetamol at poly(L-Leucine) layered carbon nanotube paste electrode, Surfaces and Interfaces 24, 2021, 101154, IF: 4.83.

9. G. Tigari, J. G. Manjunatha, **Edwin S D' Souza**, N. Sreeharsha, Surfactant and Polymer Composite Modified Electrode for the Sensitive Determination of Vanillin in Food Sample. ChemistrySelect, 6, 2021, 2700 – 2708. DOI:10.1002/slct.202004370, IF: 2.109.

10. Girish Tigari, J. G. Manjunatha, **Edwin S D' Souza**, C. Raril, N. Hareesha, M. M. Charithra. Electrochemical determination of levofloxacin drug at poly (clayton yellow)/carbon paste electrode. Chemical Monthly, https://doi.org/10.1007/s00706-022-02910-2, IF:1.6.

Papers Presented in National/International Conferences:

1. Edwin S. D' Souza, One day national seminar on E-Resources, Intellectual property rights and plagiarism-Issues and challenges, St. Philomena College, Puttur, Karnataka, 29th September, 2018.

2. Edwin S. D' Souza, J.G Manjunatha, Electrochemical studies of Dopamine and Uric Acid and their simultaneous determination at poly (Proline), International Conference on Nanotechnology - 2019, St. Aloysius college, Mangalore, 10 – 11 January, 2019.

3. Edwin S. D' Souza, J.G Manjunatha, Development of Electrochemical Sensor for the Determination of Folic Acid, National level conference, FMKMC College Madikeri, Mangalore University Constituent College, 05th February 2019.

4. Edwin S. D' Souza, J.G Manjunatha, Arginine Electropolymerized Carbon Nanotube Paste Electrode for Electrochemical Determination of Vanillin, One Day National Seminar on Current Trends in Chemistry Research and Development (CTCRD-2019), Vivekananda College of Arts, Science and Commerce, Puttur, 18th September 2019.

5. Edwin S. D' Souza, Manjunatha, J. G., Electropolymerized Threonine on Carbon Paste Electrode as Sensitive Sensor for Electrochemical Determination of Riboflavin, International Conference on Advances in Chemical & Materials Sciences (ICCM), Department of Studies in Chemistry, Mangalore University, 17-19, October 2019.

6. Edwin S. D' Souza, J. G. Manjunatha, Polymer Modified Carbon Paste Electrode for the Electrochemical Determination of Riboflavin, National level conference, FMKMC College Madikeri, Mangalore University Constituent College, 12th February 2020.

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